CALIFORNIA ENVIRONMENTAL PROTECTION AGENCY
DEPARTMENT OF PESTICIDE REGULATION
MEDICAL TOXICOLOGY BRANCH
SUMMARY OF TOXICOLOGY DATA

Nabam

Chemical Code # 000417, DPN # 50320
29 August 1991
Revised March 23,1992, January 2, 2002 and February 9, 2005

I. DATA GAP STATUS

Chronic toxicity, rat:	Data gap, no study on file[a].
Chronic toxicity, dog:	Data gap, no study on file[a]
Oncogenicity, rat:	Data gap, no study on file[a].
Oncogenicity, mouse:	Data gap, no study on file[a].
Reproduction, rat:	Data gap, no study on file[a]
Teratology, rat:	Data gap, no study on file[a].
Teratology, rabbit:	No data gap, possible adverse effect
Gene mutation:	No data gap, no adverse effect.
Chromosome effects:	No data gap, possible adverse effect.
DNA damage:	No data gap, possible adverse effect.
Neurotoxicity:	No data gap, no adverse effect.

Toxicology one-liners are attached.
All record numbers through 128865 were examined.
** indicates an acceptable study.
Bold face indicates a possible adverse effect.
Previous version by S. Morris and J. Gee, 3/23/92
File name: T050209 doc
Revised by H. Green and J. Gee, 01/02/2002, by Gee, 02/09/05
Note: EPA's "Guidance for the Reregistration of Pesticide Products Containing Nabam as the Active Ingredient" was published April 1987. See also the January 1996 RED.

[a] In a memorandum, dated May 20, 2003, the Office of Environmental Health Hazard Assessment concurred with the Medical Toxicology Branch of the Department of Pesticide Regulation that the remaining studies for nabam may be waived at this time, based on the existing studies and on data available on ETU, the major metabolite.

II. TOXICOLOGY ONE-LINERS AND CONCLUSIONS

These pages contain summaries only. Individual worksheets may contain additional effects.

50320 – 012 "Chemical Manufacturers Association antimicrobial exposure assessment study." (W. Popendorf, M. Selim and B. C. Kross, University of Iowa, 2/28/90) The study was conducted in response to the Data Call-In for antimicrobial active ingredients of March 4, 1987. Emphasis was on dermal and inhalation exposure during applications. No worksheet or detailed review. (Gee, 6/25/01)

50320 – 013 "Nabam Task Force response to the EBDC PD 2/3: Toxicity and exposure assessment." (D. J. Severn, Jellinek, Schwartz, Connolly & Freshman, Inc., 4/2/90) The brief document discusses using the Iowa study for assessment of exposure as more relevant than agricultural studies. No worksheet or detailed review. (Gee, 6/25/01).

COMBINED, RAT

No study on file.

CHRONIC TOXICITY, RAT

No study on file.

CHRONIC TOXICITY, DOG

No study on file.

ONCOGENICITY, RAT

No study on file.

ONCOGENICITY, MOUSE

No study on file.

REPRODUCTION, RAT

No study on file.

TERATOLOGY, RAT

No study on file.

TERATOLOGY, RABBIT

**** 50320-014 128865** "A Developmental Toxicity Study of Aquatreat DN-30 in Rabbits" (Mark D. Nemec, WIL Research Laboratories, Inc., Ashland OH., Report # WIL-190002, 30 July 1992). Twenty artificially inseminated female New Zealand White rabbits per group received Aquatreat DN-30 (32.79% Nabam in water) by gavage at concentrations of 0 (water), 1, 8, and 100 mg/kg/day [equivalent to 0.328, 2.62 and 32.8 mg/kg of nabam] on gestation days 7 through 19. Mean ETU content in bulk test material was 0.5 – 0.6%. Mean post-implantation loss was increased at 100 mg/kg/day compared to controls (1.8 at 100 mg/kg compared with 1.1 in controls). Group mean maternal food consumption and body weight gain were reduced during treatment at 100 mg/kg/day relative to controls. Maternal NOEL = 8 mg/kg/day. **Possible Adverse effects: the incidences of hydrocephaly and cleft palate were increased at 8 and 100 mg/kg/day relative to historical controls and to concurrent controls.** Developmental NOEL = 1 mg/kg/day. **Acceptable.** (H. Green and Gee, 6/22/01)

GENE MUTATION

** 50320-001 045553 J. Cavagnaro and R. C. Sernau, "CHO/HGPRT Forward Mutation Assay, Nabam, Final Report", Hazleton Biotechnologies Corp., Vienna, VA., project # 752-111, 9/9/85. The forward mutation rate of the HGPRT locus was analyzed by measuring the cloning efficiency of CHO cells in the presence of thioguanine after exposure to nabam (unstated purity and lot #, 30% solution) without metabolic activation at 0, 2, 4, 6, 8, or 10 µg/ml and with metabolic activation from rat liver at 0, 8, 60, 90, 100, 120, 150, or 300 µg/ml or from mouse liver at 0, 4, 6, 8, 10, or 20 µg/ml. No adverse effect was indicated. The study was unacceptable (H. Green and S. Morris, 04/24/91) but upgraded by adequate submissions of description of the test material and experimental details (S. Morris and J. Gee, 03/23/92).

50320-002 045553: This document contains a duplicate of 50320-001 045553. No worksheet was done (S. Morris, 04/24/91).

50320-005 112635: Evaluation of these data resulted in a change in study status (see Worksheet and Response, 03/23/92; S. Morris and J. Gee, 03/23/92).

** 50320-001 045557, *"Salmonella typhimurium*/Mammalian Microsome Plate Incorporation Assay with Compound Nabam, Lot # 28177DP, LH #21,952A, Final Report", Hazleton Biotechnologies Corporation, Vienna, VA., Project # 752-108, 10/7/85. Nabam (30% stock solution, lot #28177DP) was tested in a bacterial mutation assay that measured the frequency that histidine auxotrophic strains of *Salmonella typhimurium* (TA1535, TA1537, TA1538, TA100, and TA98) reverted to histidine prototrophic growth. The assays were conducted in triplicate with and without a metabolic activation system (S-9 fractions of Fischer 344 rat or B6C3F1 mouse liver homogenates) at 0, 0.3**,** 1.3, 6.7, 33.3, 100.0, or 300.0 µg/plate. There were no compound-related increases in revertant rates. No adverse effect was indicated. The study was unacceptable (H. Green

and S. Morris, 05/20/91) but upgraded by adequate submissions of: description of the test material, rationale for the doses used, description of the metabolic activation systems, viability data for each treatment group, and individual plate counts (S. Morris and J. Gee, 03/23/92).

50320-002 045557: This document contains a duplicate of 50320-001 045557. No worksheet was done (S. Morris, 05/20/91).

50320-005 112631: Evaluation of these data resulted in a change in study status (see Worksheet and Response, 03/23/92; S. Morris and J. Gee, 03/23/92).

CHROMOSOME EFFECTS

50320-001 045556 "Clastogenic Evaluation of Nabam, 30% Water Solution, Alco/Vining/Uniroyal, lot # 28177DP, in the Rat Bone Marrow Cytogenetic Assay, Amended Final Report." Litton Bionetics, Inc., Kensington, MD., project # 22202, March 1986. Nabam was given by oral gavage to 10 male Fischer 344 rats per group at 0, 80, 270, or 400 mg/kg. The rats were sacrificed at 6, 24, or 48 hours after a single acute dose or 6 hours after the last of 5 consecutive daily doses. The rats were injected ip with 2.0 mg/kg colchicine 3 hours prior to sacrifice and bone marrow cells were harvested, fixed, and stained. Fifty metaphase cells per animal were examined microscopically and scored for chromosome aberrations. No adverse effect was indicated. The study was unacceptable but possibly upgradeable with submission of adequate rationale for using only one sex (H. Green and S. Morris, 5/17/91).

50320-002 045556: This document contains a duplicate of 50320-001 045556. No worksheet was done (S. Morris, 5/17/91).

50320-005, letter dated 1/27/92: This document contains a statement of purity for the test material used in 50320-001 045556. No worksheet was done (S. Morris, 3/23/92).

** **50320-001 045558** "Clastogenic Evaluation of Nabam, 30% Water Solution, Alco/Vining/Uniroyal, lot # 28177DP, in an *In Vitro* Cytogenetic Assay Measuring Sister Chromatid Exchange in Chinese Hamster Ovary (CHO) Cells, Third Amended Final Report", LBI Project No. 20990, Litton Bionetics, Inc., Kensington, MD., April 1986. Nabam (30% stock solution) was tested for induction of sister chromatic exchanges *in vitro* in cultured Chinese Hamster ovary cells. Duplicate cultures were exposed with and without a metabolic activation system (S-9 fractions from Aroclor 1254-induced male Fischer 344 rat or male B6C3F1 mouse liver homogenates) at 0, 1.00, 1.67, 3.30, or 5.00 mg/ml or with rat S-9 at 0, 2.0, 3.0, 4.0, or 5.0 mg/ml. **A possible adverse effect was indicated by treatment-related increases in sister chromatic exchanges with and without metabolic activation at all treatment levels**. The study was upgraded from unacceptable (H. Green and S. Morris, 05/23/91) to acceptable by submission of an adequate description of the test material (S. Morris and J. Gee, 3/20/92).

50320-005, letter dated 1/27/92: Evaluation of these data resulted in a change in study status (see Worksheet, 3/20/92 and Response, 03/23/92; S. Morris and J. Gee, 03/23/92).

** 50320-005 112636 "Mutagenicity Test on Aquatreat ON-30 (30X Nabam in Water) in the Rat Bone Marrow Cytogenetic Assay." J.L. Ivett, Hazleton Laboratories America, Inc.,

Kensington, MD; ID # 10645-0-452; 7/3/89. Groups of 5 Sprague-Dawley rats / sex / time point were given Aquatreat ON-30 (30% Nabam in water, lot # 5243, water vehicle at 10 ml/kg) by oral gavage at 120, 400, or 1200 mg/kg and sacrificed 6, 18, or 30 hours later. The negative (water) and positive (60 mg/kg cyclophosphamide) control groups were sacrificed, respectively, 30 and 18 hours after oral gavage. IP injections of 2.0 mg/kg colchicine were given 2 hours before sacrifice. At sacrifice, tibia bone marrow was collected, fixed, stained, and 50 metaphase cells / rat were scored for chromosome aberrations. There were no treatment-related effects. The positive controls and dosing rationale were adequate. **No adverse effect was indicated**. The study was acceptable (S. Morris and J. Gee, 3/19/92).

DNA DAMAGE

50320-001 045554 "Evaluation of Nabam in the C3H-1OT 1/2 Cell System for Transformation and Promotion Activities." Arthur D. Little, Inc., Acorn Park, Cambridge, MA., 9/24/85. Solutions of nabam (unstated purity, lot # 28177 DP, 30% stock solution) were tested on cultured C3H-1OT 1/2 mouse embryo fibroblasts. Preliminary toxicity data indicated plating efficiency was decreased ~ 99% by treatment with 1.0 ng/ml test material in the absence of metabolic activation and - 47% by treatment with 100 ng/ml in the presence of metabolic activation (S-9 fraction of Aroclor 1254 induced Sprague-Dawley rat liver homogenate). Treatment without activation at 0, 0.1, 0.2, 0.4, 0.6, or 0.8 µg/ml; with activation at 0, 25, 50, 100, or 200 µg/ml; or a 24-hour treatment with 0.5 µg/ml MNNG followed by a continuous exposure to 0.05 µg/ml of test material for ≈ 5 weeks produced no treatment-related increases in transformed foci. **No adverse effect was indicated**. The results of the promotion assay should not be used to characterize toxicity. The study is unacceptable but possibly upgradeable with submission of adequate analysis of exposure solutions (H. Green and S. Morris, 05/07/91).

50320-001 045555 This is the same document as CDFA doc. # 50320-001 rec. # 045554. A separate worksheet was not done (S. Morris, 05/07/91).

50320-002 045555 This document contains a duplicate of 50320-001 045554. No worksheet was done (S. Morris, 05/07/91).

** **50320-001 045559** "Unscheduled DNA Synthesis Rat Hepatocyte Assay with Nabam, Final Report." Project No. 752-110, Hazleton Biotechnologies Corporation, Vienna, VA., 9/17/85. Nabam (30 % stock solution) was tested for cytotoxicity and unscheduled DNA synthesis (UDS) at 0 (water), 0.1, 0.5, 1.0, 5.0, 10, 50, or 100 µg/ml by exposing primary hepatocytes from a male Fischer 344 rat "overnight" to the test material in the presence of [3H] Thymidine. Cytotoxicity was measured by trypan blue exclusion. UDS was analyzed by autoradiographic measurement of nuclear incorporation of ^{3}H. Cytotoxicity was observed at doses $\geq$10 µg/ml (33% survival). **A possible adverse effect was indicated by an increase in net nuclear grain counts at 5.0 µg/ml**. The study was unacceptable (H. Green and S. Morris, 05/31/91) but upgraded with an adequate description of the test material (S. Morris and J. Gee, 3/23/92).

50320-005, letter dated 1/27/92: Evaluation of these data resulted in a change in study status (see Worksheet and Response, 03/23/92. (S. Morris and J. Gee, 03/23/92).

50320-005 112637 "Mutagenicity Test on Aquatreat DN-30 (30X Nabam in water) in the *In*

Vitro Transformation of BALB/C-3T3 Cells Assay with an S9 Activation System." HLA Study No: 10645-0-488; B.C. Myhr; Hazleton Laboratories America, Inc., Kensington, Maryland. Nabam (Aquatreat DN-30, lot #5243) was assayed for cellular transformation in 8 graded doses (made up in growth medium) from 12 to 288 µg/ml (trial 1) and 12 to 184 µg/ml (trial 2). Twenty-five cm^2 culture flasks were seeded with 3 x 10^4 cells, incubated for one day, then exposed to the test material for four hours in the presence of S9 metabolic activation system from liver homogenates of Aroclor 1254-induced male Sprague-Dawley rats. The cultures were then washed and incubated in growth medium for four weeks and then scored for non-contact-inhibition foci / culture. **No adverse effect was indicated**. The study was unacceptable and not upgradeable because there were no trials without metabolic activation and the assay lacked sensitivity to positive controls (S. Morris and J. Gee, 3/12/92).

NEUROTOXICITY

** 50320-008 126321 "A Combined Subchronic (13 Week) Toxicity and Neurotoxicity Study of Aquatreat DN-30 in Rats." (Ian C. Lamb, WIL Research Laboratories, Inc., Ashland, Ohio, Report # WIL-190004, 12 April 1993). Ten (10) or 20 Sprague-Dawley, Crl:CD®BR rats per sex per group received Aquatreat DN-30 (31.1% and 32.8% Nabam) by gavage at 0 (deionized water), 0.08 (10 animals per sex), 0.80, 80, and 260 mg/kg/day (0, 0.0262, 0.2623, 26.232, and 85.25 mg/kg/day Nabam respectively) for at least 91 consecutive days. ETU content was 5527 to 6226 µg/ml of the bulk material. Ten animals per sex per group were used for subchronic neurotoxicity evaluation and the remainder were used for general subchronic toxicity evaluation. All animals in the low dose (0.08 mg/kg/day) were used for subchronic toxicity evaluation. Functional Observational Battery (FOB) and motor activity results were unremarkable at weeks 4, 8 and 13 compared with pretest values. Absolute and relative thyroid gland weights were increased at 260 mg/kg for males and females and at 80 mg/kg/day for females . Thyroxine (T4) was statistically lower in both sexes at 80 and 260 mg/kg. TSH was significantly increased in high dose males at weeks 6 and 13. Although TSH was also increased in females at the high dose, the increase was not statistically significant. In the subchronic toxicity animals, there was an increased incidence of hypertrophy of the thyroid follicular epithelium for high dose males (3/10) compared with controls (0/10); no hypertrophy was reported for females. No microscopic changes were recorded for the perfused neurotoxicity group animals (5/sex in controls and high dose groups). Neurotoxicity NOEL = 260 mg/kg/day. Subchronic NOEL = 0.8 mg/kg/day (reduced thyroxine (T4) levels in both sexes at 80 and 260 mg/kg/day, increased thyroid gland weights in females). **No adverse effects were indicated. Acceptable.** (H. Green and J. Gee, 6/26/01).

50320 – 009 126322 "A 21-Day Range Finding Acute Study of Aquatreat DN-30 in Rats" (Ian C. Lamb, WIL Research Laboratories, Inc., Ashland, OH., Report # WIL-190003, 5 May 1993). Five Sprague-Dawley Crl:CD®BR rats per sex per group received Aquatreat DN-30 (32.79% Nabam, lot # 23409) by gavage at concentrations of 0 (deionized water), 0.1, 1.0, 10.0, 100, and 500 mg/kg/day (0, 0.033, 0.33, 3.28, 32.79, and 163.95 mg/kg/day active Nabam, respectively) for 21 consecutive days. ETU content was 5200 to 6200 µg/ml. Weekly bodyweight and bodyweight gains were reduced at 100 (females) and 500 mg/kg/day (both sexes) compared to controls (not statistically significant). Decreases in thyroxine (T4) were noted at 100 (females) and 500 mg/kg/day (both sexes) relative to controls. Thyroid stimulating hormone (TSH) levels were increased at 500 mg/kg/day for both sexes. Increased thyroid gland weights

were recorded at 100 (males) and 500 mg/kg/day (both sexes). Hypertrophy of the thyroid follicular epithelium was observed at 100 (minimal) and 500 (minimal to mild) mg/kg/day in males and females. Apparent NOEL = 10 mg/kg/day. No worksheet. Supplemental data. (Gee, 6/26/01)

SUPPLEMENTAL

50320-001 045547 Graham et al. (1973), "Effects of one-year administration of ethylenethiourea upon the thyroid of the rat," J. Agr. Food Chem., 21 (3):324-329. This document contained an article from the open literature that reported a study conducted with a possible contaminant of nabam. No worksheet was done (S. Morris, 04/24/91).

152-013 020970: This document contains a duplicate of 50320-001 045547. No worksheet was done (S. Morris, 04/24/91).

50320-002 045547: This document contains a duplicate of 50320-001 045547. No worksheet was done (S. Morris, 04/24/91).

50320-001 045548, Graham et al. (1975), Effects of prolonged ethylene thiourea ingestion on the thyroid of the rat, Fd. Cosmet. Toxicol., 13:493-499. This document contained an article from the open literature that reported a study conducted with a possible contaminant of nabam. No worksheet was done (S. Morris, 04/24/91).

152-013 020971: This document contains a duplicate of 50320-001 045548. No worksheet was done (S. Morris, 04/24/91).

50320-002 045548: This document contains a duplicate of 50320-001 045548. No worksheet was done (S. Morris, 04/24/91).

** 50320 - 010 126323 “Twenty-One Day Sub-Chronic Dermal Toxicity Study with Aquatreat DN-30 (approximately 30% Nabam in Water) in Rats” (M.E.P Goad, Arthur D. Little, Inc., Cambridge, MA, Report # 41178, 11 May 1993). Clipped, unabraided skin (10% of body surface) of five (5) Sprague-Dawley rats per sex per group was treated with Aquatreat DN-30 (32.8% Nabam, lot # 23409) at 0, 46, 457, 1525, and 3050 mg/kg (equivalent to 0, 15, 150, 500, and 1000 mg/kg Nabam, respectively) for 6 hours per day, 5 days per week for 3 weeks. ETU content was not evaluated. Test material was applied to gauze, which was then placed on the skin, since the test material rolled off into the fur when applied directly to the skin (due to the volume used). The gauze patches were held in place with tape. After the treatment, sites were washed. Males and females at 457 (150 Nabam), 1525 (500 Nabam), and 3050 (1000 Nabam) mg/kg developed slight to moderate erythema at the test site. Incidences were 0, 0, 1, 3 and 5 for males with increasing dose and 0, 0, 2, 1 and 3 for females. There were no systemic effects on body weight, food consumption, hematology, clinical chemistry or histopathology. Thyroid organ weight was comparable among groups. Measurements of thyroid hormones T3, T4 and TSH showed some variation with statistically significant differences in females for T3 and T4. These differences were explained as due to the order of sampling of animals for blood. Thyroid hormones show diurnal variability and animals were sampled from Group 1 males through Group 5 of females (last), with the peak levels occurring at night. All levels, however, were within or

close to normal values. Dermal NOEL = 46 mg/kg Aquatreat DN-30 (15 mg/kg Nabam). Systemic NOEL = 3050 mg/kg (1000 mg/kg Nabam). **No Adverse effects. Acceptable.** (H. Green and Gee, 6/27/01).